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ECBC-TR-593

EVALUATION OF VIKING 573 GC/MS SYSTEM AND CHEMICAL WARFARE AGENT (CWA) DETECTION

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PREFACE

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CONTENTS

1.	INTRODUCTION	7
2.	REQUIRED EQUIPMENT AND MATERIALS.....	7
2.1	Equipment	7
2.2	Materials	8
3.	METHODOLOGY AND PROCEDURES.....	8
3.1	GC/MSD Technology Introduction	8
3.2	Methodology and Data Interpretation.....	10
3.3	Sample Preparation Information	11
3.3.1	Organic Matrix Sample Preparation	11
3.3.2	Soil Matrix Sample Preparation and Extraction	12
4.	RESULTS AND DISCUSSIONS.....	13
4.1	LOD Results.....	13
4.2	Dynamic Range Results.....	14
5.	CONCLUSIONS.....	15
	APPENDIX - SUPPORTING DATA.....	17

FIGURES

1.	GC and EI-quadrupole MS Operation	9
2a.	20 mL I-Chem Vial, Syringe Needle, 10-mL Syringe, and Gelman Acrodisc 0.45- μ m Syringe Filter.....	13
2b.	Syringe and Needle Configuration.....	13
2c.	Syringe and Filter Configuration	13
A-1	Instrument performance check with 1 ppm Hexachlorobenzene standard	17
A-2	GA Standard Curve.....	20
A-3	GB Standard Curve	20
A-4	GD Standard Curve.....	20
A-5	GF Standard Curve	20
A-6	VX Standard Curve.....	21
A-7	HD Standard Curve.....	21

TABLES

1.	CWA Standards Preparation	11
2.	Soil Sample Extraction Preparation.....	12
3.	Estimated LODs of CWA in Solvent.....	13
4.	Estimated LODs of CWA in Soil Matrix.....	14
5.	Dynamic Range of CWA Standards for Viking GC/MS 573 System	14
A-1	Library Match and S/N Data Summary for LODs of CWA in Solvent.....	18
A-2	Data Summary for CWA in Soil Matrix	18
A-3	Dynamic Range Data for CWA Standards	19

EVALUATION OF VIKING 573 GC/MS SYSTEM AND CHEMICAL WARFARE AGENT (CWA) DETECTION

1. INTRODUCTION

The New York City Department of Environmental Protection (NYCDEP), Division of Emergency Response and Technical Assessment (DERTA) funded the Mobile Laboratories & Kits (ML&K) Team, Edgewood Chemical Biological Center (ECBC), to evaluate a portable Gas Chromatography/Mass Spectrometry (GC/MS) system that they routinely employ in mobile laboratory applications. A Bruker Viking 573 GC/MS system was provided by the NYCDEP DERTA. The instrument were verified according to manufacturer's recommendations, and members of the ML&K Team developed methods to evaluate the capabilities of the system against six chemical warfare agents (CWAs) including the Limit of Detection (LOD) and dynamic range for all six CWA compounds in organic solvent and soil matrices.

2. REQUIRED EQUIPMENT AND MATERIALS

2.1 Equipment

- Bruker Viking 573 GC/MS system
- Chemical fume hood
- Analytical pipettes
- Analytical balance (minimum ± 0.01 mg accuracy)
- Hamilton syringes
- 10mL disposal plastic syringes
- 20mL I-Chem vials
- Disposable plastic syringe needle
- Gelman Acrodisc 0.45 μ m syringe filter
- Stainless steel spatulas
- Auto-sampler vial crimper and de-crimper

2.2

Materials

- 1 mL x 100 µg/mL stock combo solution of GA, GB, GD, GF, and HD in methylene chloride
- 1 mL x 100 µg/mL stock solution of VX in Isopropyl Alcohol (IPA)
- 1mL x 150 µg/mL stock combo solution of GA, GB, GD, GF, HD, and VX in methylene chloride
- Auto-sampler vials
- Methylene chloride (CH₂Cl₂)
- Isopropyl alcohol (IPA)
- Soil
- Kim-wipes
- Nitrile gloves
- 2 mL auto-sampler vials
- Disposable pipette tips

3.

METHODOLOGY AND PROCEDURES

The ML&K Team chemists and technicians developed methods to perform validation testing specifically on a Bruker Viking Spectratrak 573 GC/MS system owned by NYCDEP. Six parent chemical warfare agents (CWAs) to include GA, GB, GD, GF, HD and VX, were to be evaluated to determine the detection and identification capabilities of the instrument. The Limits of Detection (LODs) and dynamic ranges were determined for each analyte. These parameters were evaluated on standard samples in a soil and organic solvent matrix. The soil was spiked with a reasonable level of each analyte, extracted with the appropriate solvent, and the sample and sample extract was introduced or injected into the instrument using manual injection techniques and then analyzed. All sample injections were made manually and all work was performed in accordance with the stringent requirements of the Edgewood Chemical/Biological Forensic Analytical Center's Quality System.

3.1

GC/MSD Technology Introduction

GC/MSD couples a gas phase separation technique with Mass Selective Detection to provide a highly sensitive and specific detection and identification method for relatively non-polar volatile and semi-volatile molecules. Current GCs are composed of a temperature

programmable injection port interfaced with an analytical capillary column housed in a temperature programmable heated oven. An inert gas such as helium serves as the mobile phase or carrier gas, but hydrogen, which is not inert, can also serve as the mobile phase or carrier gas. The column is lined with a liquid stationary phase. The injection port is held at a temperature well above the boiling point of any target analyte. Upon injection, the target analyte flash vaporizes and is subsequently swept into the analytical column by mobile phase. The head of the column is initially held at a temperature well below any target analyte's boiling point, causing analytes to immediately condense at the head of the column. The column is then subjected to a temperature program. As respective analyte boiling points are reached, they re-enter the gas phase and are swept through the column. Separation of complex mixtures occurs due to characteristic partitioning of target analytes between mobile and stationary phases. Upon elution from the analytical column, analytes are transferred from the relative ambient pressure of the GC to the high vacuum conditions of the mass spectrometer.

A mass spectrometer consists of an ion source, ion focusing optics, a mass filter, and a detector. The instrument to be evaluated employs an Electron Ionization (EI) source to create ions and a quadrupole mass filter to isolate and collect them as shown in Figure 1.

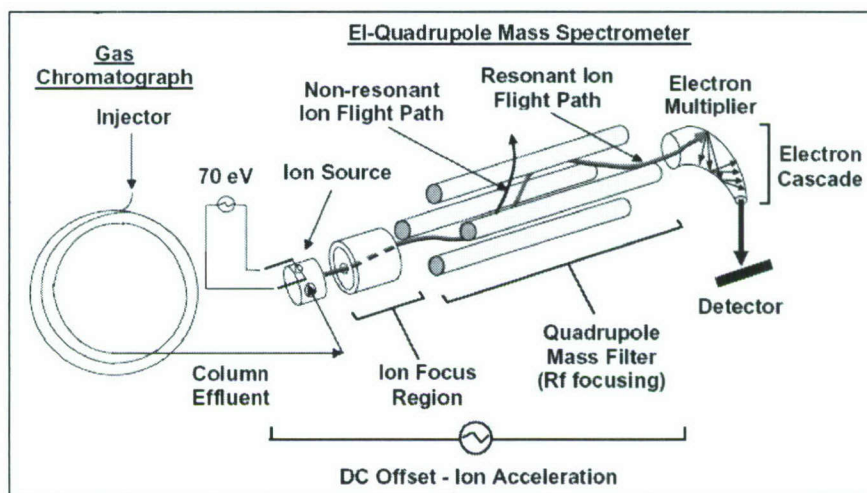


Figure 1. GC and EI-quadrupole MS Operation

Upon entering the source region, energy is transferred to analytes by a high-energy electron beam to produce charged species or ions. Ions are created when an electron from the beam knocks an electron off of analyte atoms. Ions are then accelerated and focused by the direct current offsets and radio frequency (rf) fields, respectively, through an optic focusing region into the mass filter. In a quadrupole mass filter, ions are subjected to rf electromagnetic fields to separate them in time based on their mass-to-charge ratio (m/z). Opposing quadrupole rods cycle through rf frequencies out of phase with one another causing resonant ions to fly to the detector, whereas, non-resonant ions and neutral species are removed by the vacuum system.

The ion separation power of a mass spectrometer is described by the resolution, which is defined as:

$$R = m / m'$$

where m is the ion mass and m' is the difference in mass between two resolvable peaks in a mass spectrum (e.g., a mass spectrometer with a resolution of 1000 can resolve an ion with an m/z of 100.0 from an ion with an m/z of 100.1).

3.2 Methodology and Data Interpretation

The GC/MSD system is outfitted with a 30 m x 0.25 mm ID (0.25 μ m film thickness, DB5-MS liquid phase) capillary GC column. The system is interfaced with a bottle of grade V-He to supply high purity carrier gas. To determine chromatographic characteristics of GA, GB, GD, GF, HD and VX, a 1 μ L manual injection of a standard containing 50 ng of each analyte/ μ L methylene chloride was analyzed. GC parameters were programmed as follows:

- Initial Oven Temp: 40°C
- Injector Temp: 250°C
- Pressure/Flow Mode: Constant Flow-1.2 to 1.5 mL/min
- Split Ratio Flow: 36.1 mL/min
- Carrier gas: He
- Injection volume: 1 μ L
- Solvents: CH₂Cl₂ and Isopropyl Alcohol (IPA)
- Oven Ramping:

Ramp	Rate (°C/min)	Final Temp. (°C)	Final Time (min)
1	0	40	2.00
2	10	160	1.00
3	20	280	5.00
Run Time			26.00

Prior to the initial weekly analysis and on an “as needed” basis, the Mass Selective Detector (MSD) will be “standard spectrum” auto-tuned (Target Tune) with Perfluorotributylamine (PFTBA). The mass assignments, mass resolution, ion ratios, and isotopic ratios will be evaluated and must meet the following criteria:

- Mass assignment: within ± 0.20 amu

- Mass peak width at half peak height: 0.50 ± 0.10 amu
- Ion ratios: base peak-69; relative abundance for 219 m/z >40% and 502 m/z >2%
- Isotopic ratios: 0.5-1.5%, 2-8% and 5-15% for masses 70, 220, and 503, respectively (1, 5 and 9% theoretical)

In addition to auto-tune, instrument performance was also checked with 1 ppm Hexachlorobenzene (HCB) standard. HCB standard data was obtained daily before analyzing any samples. The results are shown in Table A-1 of the Appendix. The performance specification for 1ppm HCB standard was

- Full scan NIST (National Institute of Standards and Technology) spectral library match or Automated Mass Spectral Deconvolution and Identification System (AMDIS) search must be >90%
- Full scan signal-to-noise (S/N) ratio must be $\geq 1000:1$

During analyses, the MSD will be run in scan mode over a mass range of 40-450 amu. Requirements for LOD determination include the following:

- Full scan NIST spectral library match or AMDIS search must be >80%
- Full scan S/N must be $\geq 30:1$
- Criteria must be met for 3/3 replicates to estimate LODs

3.3 Sample Preparation Information

3.3.1 Organic Matrix Sample Preparation

Single and combo chemical warfare agents (CWA) standards were prepared at concentrations indicated in Table 1. Methylene chloride was used as a solvent for GA, GB, GD, GF, and HD. Isopropyl alcohol (IPA) was used for VX since earlier stability studies have concluded that VX is more stable in isopropyl alcohol.

Table 1. CWA Standards Preparation

CWA Analytes	Single/Combo Concentrations [µg/mL]						
GA	1	5	10	25	50	110	150
GB	1	5	10	25	50	100	
GD							
GF							
HD							
VX	1	5	10	25	50	100	

3.3.2 Soil Matrix Sample Preparation and Extraction

Soil extraction samples were prepared as indicated in Table 2 using the 5-steps method described below.

Table 2. Soil Sample Extraction Preparation

CWA Analyte in the mix	Stock CWA Mix [µg/mL]	Spiked Vol. of Stock CWA Mix (mL)	Amount of Soil Matrix (g)	Vol. of MeCl (mL)	Calculated concentration of CWA mix/soil extract (µg/mL)
GA	25	1.0	1.0	3.0	6.25
GB					
GD					
GF					
VX					
HD					

Step 1:

Prepare 2.0 mL of 25 µg/mL CWA mix that contains GA, GB, GD, GF, VX, and HD in methylene chloride, as indicated in Table 2

Step 2:

Spike 1.0 g of soil in a 20-mL I-Chem sample vial with 1 mL of 25 µg/mL CWA mix solution

Step 3:

Add 3.0 mL of methylene chloride into the spiked CWA/soil sample vial and tightly close the cap

Step 4:

Carefully hold the sample vial and shake it vigorously for 30 s. Gently tap the bottom of the vial onto the palm of your hand or onto the inside base-floor of the fume hood 3-5 times so that materials drop off the lid/cap. Allow the solution to settle until liquid and solid separate

Step 5:

Use a single-use 10-mL syringe with needle (Figure 2b) to take up 1.5 – 2.0 mL of liquid sample from the I-Chem vial. Carefully remove the needle from the syringe using a small folded Kim-wipe. Install a filter onto the syringe (Figure 2c). Carefully push the liquid through the filter and into a 2-mL GC vial. Cap and label the GC sample vial to reflect the spiking chemical name, matrix type, and calculated concentration

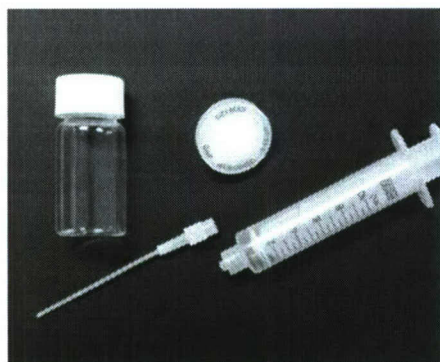


Figure 2a. 20-mL I-Chem Vial, Syringe Needle, 10 mL Syringe, and Gelman Acrodisc 0.45- μ m Syringe Filter



Figure 2b. Syringe and Needle Configuration

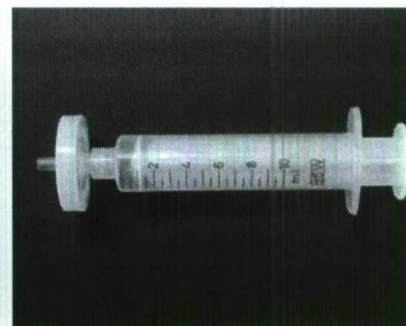


Figure 2c. Syringe and Filter Configuration

4. RESULTS AND DISCUSSIONS

4.1 LOD Results

Results are summarized in this section and the supporting data is provided in the Appendix. As shown in Table 3, the estimated limit of detections of CWA in solvent for the Viking GC/MS 573 system was determined to be 1 μ g/mL for GA, GB, GF, and HD analytes and 5 μ g/mL for GD and VX analytes with at least 95% confidence (also see Table A-1 of the Appendix for individual data).

Table 3. Estimated LODs of CWA in Solvent

CWA	LOD (μ g/mL)	Lib. Match %					S/N				
		Avg.	Stdev	CV %	C.R (90%)	C.R (95%)	Avg.	Stdev	CV %	C.R (90%)	C.R (95%)
GA	1	94.41	0.91	0.96	94.41 ± 0.33	94.41 ± 0.40	60.91	6.58	10.81	60.91 ± 2.05	60.91 ± 2.47
GB	1	95.73	2.19	2.28	95.73 ± 0.80	95.73 ± 0.97	42.32	7.46	17.63	42.32 ± 2.74	42.32 ± 3.31
GD	5	92.36	0.49	0.53	92.36 ± 0.18	92.36 ± 0.22	92.14	6.32	6.86	92.14 ± 2.32	92.14 ± 2.80
GF	1	91.18	3.22	3.53	91.18 ± 1.18	91.18 ± 1.43	46.09	11	23.87	46.09 ± 4.03	46.09 ± 4.88
VX	5	83	2.14	2.58	83 ± 0.78	83 ± 0.95	111.86	56.54	50.54	111.86 ± 20.73	111.86 ± 25.07
HD	1	93.64	1.09	1.17	93.64 ± 0.40	93.64 ± 0.48	56.73	9.03	15.92	56.73 ± 3.31	56.73 ± 4.00

Note: CV is the coefficient of variation and C.R. is the confidence range

The reported LODs met the required criteria defined in Section 3.2 as follows:

- Full scan NIST spectral library match or AMDIS search must be $>80\%$
- Full scan S/N must be $\geq 30:1$

Table 4 shows the estimated LODs of CWA in soil matrix to be 6.25 µg/mL for all testing analytes with at least 95% confidence (also see Table A-2 of the Appendix for individual data).

Table 4. Estimated LODs of CWA in Soil Matrix

CWA	LOD (ug/mL)	Lib. Match %					S/N				
		Avg.	Stdev	CV %	C.R (90%)	C.R (95%)	Avg.	Stdev	CV %	C.R (90%)	C.R (95%)
GA	6.25	96.5	2.12	2.2	96.5 ±0.78	96.5 ±0.94	41	15.56	37.94	41 ±5.71	41 ±6.90
GB		98.82	0.85	0.86	98.82 ±0.31	98.82 ±0.38	137	11.53	8.41	137 ±4.23	137 ±5.11
GD		92.55	0.51	0.55	92.55 ±0.19	92.55 ±0.23	111.77	4.85	4.34	111.77 ±1.78	111.77 ±2.15
GF		98.59	0.5	0.51	98.59 ±0.18	98.59 ±0.22	149.95	8.21	5.47	149.95 ±3.01	149.95 ±3.64
VX		84.32	0.48	0.57	84.32 ±0.18	84.32 ±0.21	214.68	20	9.31	214.68 ±7.33	214.68 ±8.87
HD		93.05	0.21	0.23	93.05 ±0.08	93.05 ±0.09	231.09	10.19	4.41	231.09 ±3.74	231.09 ±4.52

Note: CV is the coefficient of variation and C.R. is the confidence range

The reported average values for library match % and S/N variables in Tables 3 and 4 were obtained from 22 replicates. The confidence range values, C.R., at 95% confidence were also calculated indicating the library match % and S/N values at the estimated LODs of tested CWA in solvents and soil matrices met or exceeded the required criteria as mentioned above.

4.2 Dynamic Range Results

In addition to the LODs estimation, dynamic range of CWA standards for Viking GC/MS 573 system were determined as indicated in Table 5. Indeed, the MSD started getting saturated at 150 µg/mL concentration for these CWA analytes (see Table A-3 and Figures A-2 through A-7).

Table 5. Dynamic Range of CWA Standards for Viking GC/MS 573 System

Agent Names	LODs [µg/mL]	Concentration [µg/mL] that saturated detector
GA	1.0	>150
GB	1.0	≥150
GD	5.0	≥150
GF	1.0	≥150
HD	1.0	≥150
VX	5.0	≥150

5. CONCLUSIONS

The GC/MS CWA analytical method was successfully developed for the portable Viking GC/MS 573 system as indicated in Section 3.2. This method was used for determining the estimated LODs of CWA in solvents and soil matrices. At 95% confidence, the estimated LODs for CWA in solvents were reported to be 1.0 µg/mL for GA, GB, GF, and HD, and 5.0 µg/mL for GD and VX analytes. Likewise, the estimated LODs for CWA in soil matrix were determined to be 6.25 µg/mL. Dynamic range was found to be from the LOD's to ≥ 150 µg/mL.

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APPENDIX

SUPPORTING DATA

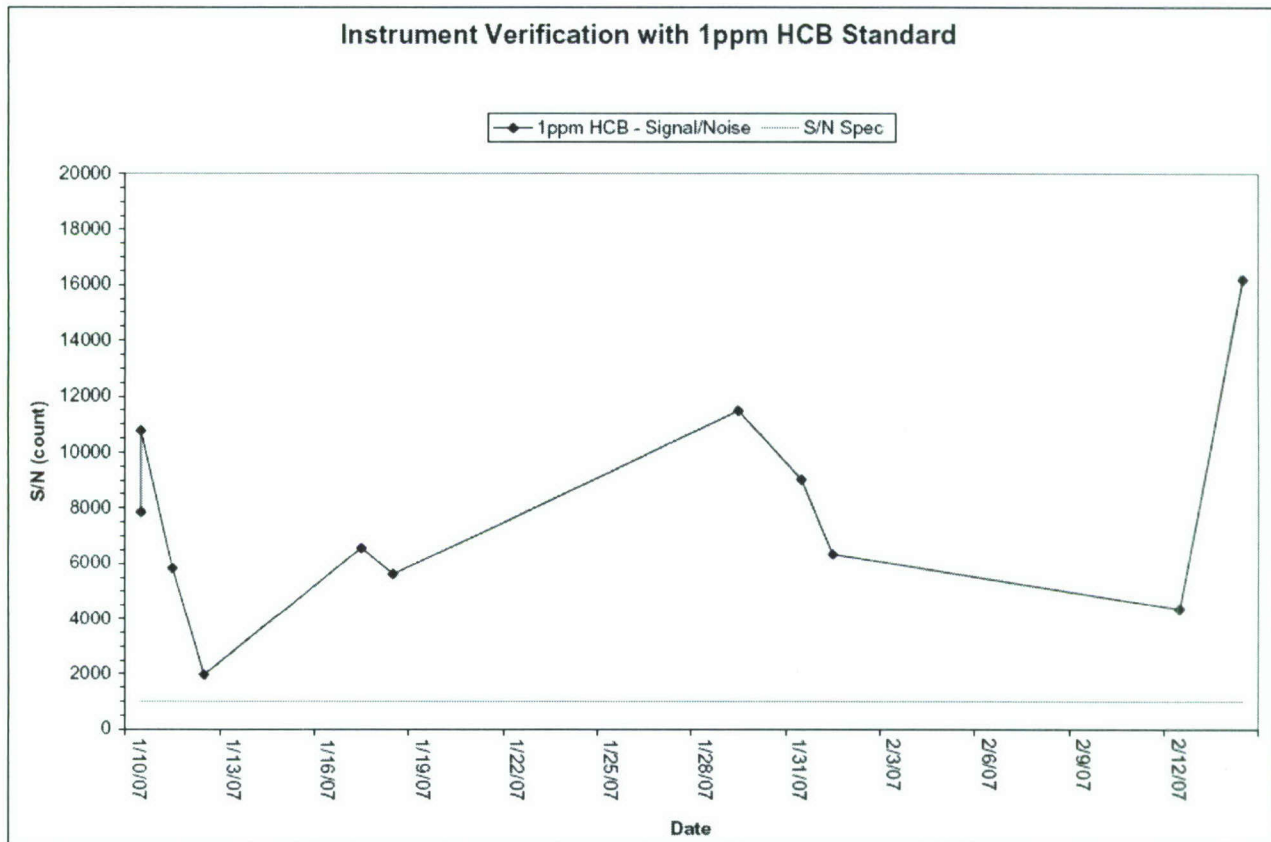


Figure A-1. Instrument performance check with 1 ppm Hexachlorobenzene standard
(Signal-to-Noise ratio of 1ppm HCB standard must be ≥ 1000 counts)

Table A-1. Library Match and S/N Data Summary for LODs of CWA in Solvent

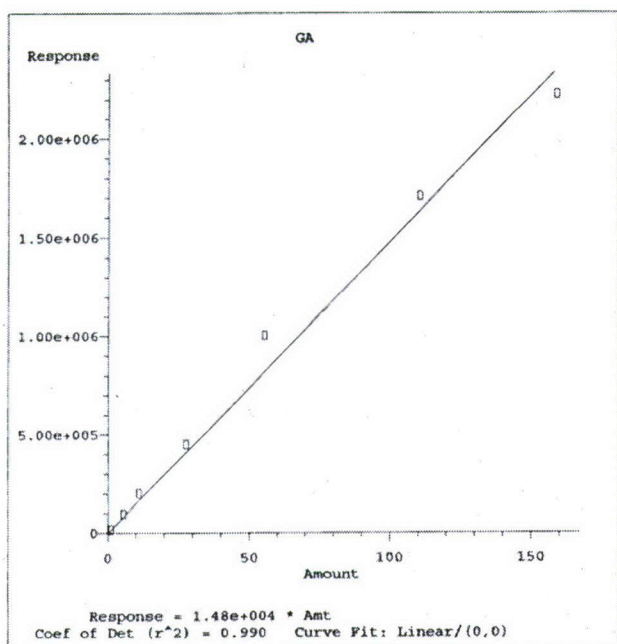
Agent Name		Replicate # and [ug/mL]	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	Mean	Stdev	CV (%)
GA	Lib. Match (%)	1	96	95	95	94	94	94	95	93	94	94	95	92	94	94	95	95	95	95	95	95	93	95	94.41	0.91	0.96
		5	83	82	87	98	91	96	96	96	84	93	92	92	93	94	98	96	85	81	84	98	75	91	90.23	6.56	7.27
	S/N	1	82	60	64	64	54	62	61	59	58	60	60	54	53	62	59	49	62	67	69	64	58	59	60.91	6.58	10.81
		5	34	42	43	38	42	42	41	59	52	59	64	53	55	66	37	56	41	41	38	50	48	49	47.27	8.47	17.92
GB	Lib. Match (%)	1	95	97	98	96	96	95	87	95	94	97	96	97	96	96	97	97	95	97	97	97	96	95	95.73	2.19	2.28
		5	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99.00	0.00	0.00
	S/N	1	37	60	49	43	41	40	22	36	40	40	49	48	42	45	49	51	45	40	41	42	35	36	42.32	7.46	17.63
		5	125	144	164	111	127	152	137	146	149	139	143	163	165	164	149	143	117	125	130	142	142	143	141.36	14.47	10.24
GD	Lib. Match (%)	1	89	89	87	83	86	84	90	80	83	85	86	82	81	82	79	81	82	82	81	80	78	80	83.18	3.40	4.09
		5	92	92	93	92	93	93	93	92	92	92	93	93	92	93	92	92	93	92	92	92	92	92	92.36	0.49	0.53
	S/N	1	23	37	27	31	27	24	44	24	26	23	30	27	29	26	25	25	29	20	21	15	23	22	26.27	5.94	22.60
		5	90	94	102	82	87	97	91	95	95	89	97	92	99	106	84	90	88	80	87	91	94	97	92.14	6.32	6.86
HD	Lib. Match (%)	1	93	93	95	93	94	94	93	94	94	94	96	92	94	94	91	95	94	93	93	95	93	93	93.64	1.09	1.17
		5	92	92	91	93	93	93	93	94	91	93	94	93	93	94	93	91	93	94	94	94	93	93	92.91	0.97	1.05
	S/N	1	66	75	65	70	64	56	35	55	54	51	64	61	60	63	55	55	57	50	48	48	47	49	56.73	9.03	15.92
		5	177	192	199	166	180	194	177	161	188	165	187	181	191	187	161	187	168	157	157	177	173	172	177.14	12.61	7.12
GF	Lib. Match (%)	1	93	96	95	94	95	94	98	88	89	92	93	89	90	91	86	88	90	88	88	92	88	89	91.18	3.22	3.53
		5	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99.00	0.00	0.00
	S/N	1	51	56	50	50	54	46	87	36	41	45	48	42	41	44	45	47	48	37	38	38	37	33	46.09	11.00	23.87
		5	152	154	161	168	167	170	158	172	179	176	176	163	166	181	161	176	144	154	161	170	151	171	164.95	10.01	6.07
VX	Lib. Match (%)	1	ND																								
		5	81	83	81	82	81	80	81	81	84	82	82	84	82	81	81	86	86	86	86	85	85	86	83.00	2.14	2.58
	S/N	1	ND																								
		5	68	79	77	79	70	73	78	65	90	79	77	83	66	65	69	184	186	187	215	193	187	191	111.86	56.54	50.54

Table A-2. Data Summary for CWA in Soil Matrix

Analytes Name	Matrix	Concentration [ug/mL]	Replicate #	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	Mean	Stdev	CV (%)	
GA	Soil/MoCl Ext.	6.25	Lib. Match %	95	92	92	90	88	95	98	89	90	92	98	98	92	96	91	91	90	91	87	91	91	94	96.50	2.12	2.20	
			S/N	42	35	36	32	32	30	52	38	34	30	30	32	30	39	35	32	33	36	35	30	33	34	41.00	15.56	37.94	
GB			Lib. Match %	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	99	95	99	99	98.82	0.85	0.86
			S/N	144	119	143	131	146	137	132	130	144	145	144	148	139	147	149	146	125	135	143	100	133	134	137.00	11.53	8.41	
GD			Lib. Match %	93	93	93	92	93	92	93	92	93	92	93	92	93	92	93	93	92	93	93	92	93	93	92	92.55	0.51	0.55
			S/N	111	105	110	111	113	115	113	115	110	109	112	112	108	117	118	124	105	114	116	111	102	108	111.77	4.85	4.34	
GF			Lib. Match %	99	99	99	99	98	99	99	99	99	98	99	99	99	99	98	98	98	99	98	99	98	98	98	98.59	0.50	0.51
			S/N	141	138	146	144	156	147	152	136	147	155	150	150	153	163	161	166	138	158	159	147	144	149	149.95	8.21	5.47	
HD			Lib. Match %	93	93	93	93	93	93	93	93	93	93	93	93	93	93	93	93	93	94	93	93	93	93	93	93.05	0.21	0.23
			S/N	229	228	241	212	228	238	217	228	242	242	243	243	233	229	221	246	226	216	229	245	220	228	231.09	10.19	4.41	
VX			Lib. Match %	84	85	85	85	85	84	84	84	84	84	85	84	84	84	85	84	84	84	84	84	85	84	84	84.32	0.48	0.57
			S/N	165	165	209	198	219	209	202	210	220	220	211	217	219	236	234	247	210	227	226	235	214	230	214.68	20.00	9.31	

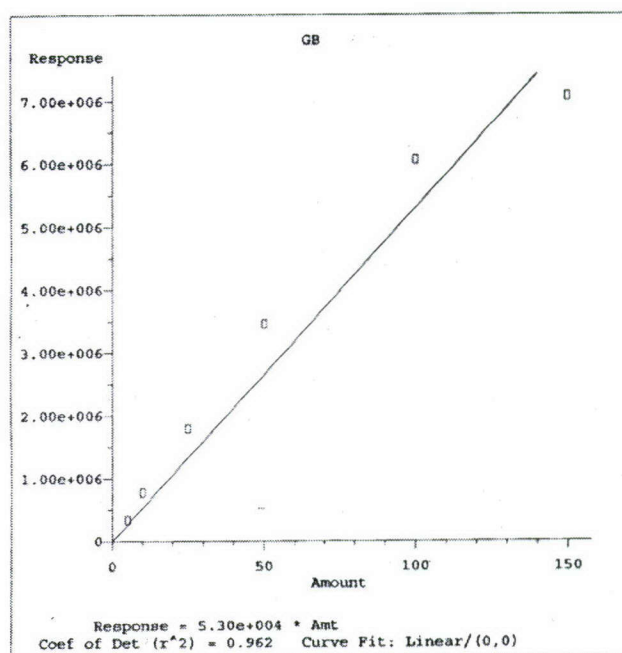
Table A-3 Dynamic Range Data for CWA Standards

Agent Name	[ug/mL]	Lib. Match %	S/N	Agent Name	[ug/mL]	Lib. Match %	S/N
GA	1	95	68	GF	1	93	51
	5	97	150		5	99	152
	10	97	249		10	98	260
	25	96	318		25	98	428
	50	96	492		50	99	564
	110	96	527		100	98	671
	150	88	180		150	98	630
GB	1	95	37	HD	1	93	66
	5	99	125		5	92	177
	10	99	239		10	93	265
	25	99	376		25	92	429
	50	99	474		50	91	511
	100	99	535		100	92	668
	150	99	536		150	93	599
GD	1	89	23	VX	1	ND	ND
	5	92	90		5	81	68
	10	93	148		10	86	222
	25	93	238		25	84	482
	50	93	324		50	82	696
	100	93	432		100	80	739
	150	93	441		150	83	687



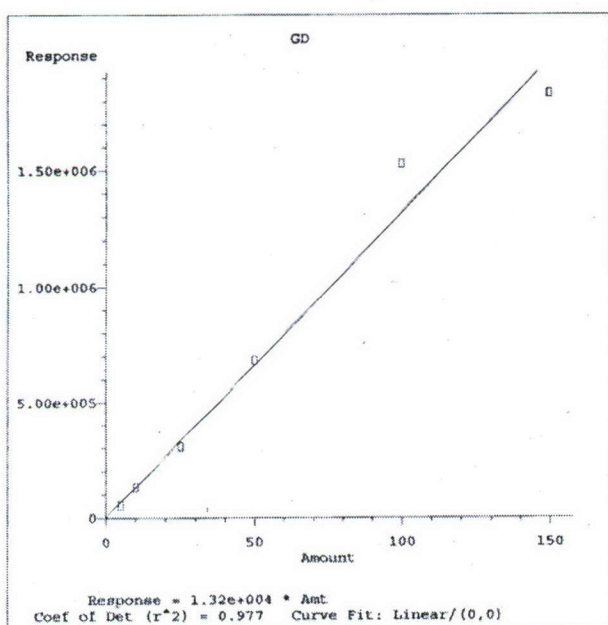
Method Name: C:\HPCHEM\METHODS\ENH\OPCWGA.M
 Calibration Table Last Updated: Fri Feb 09 17:49:38 2007

Figure A-2. GA Standard Curve



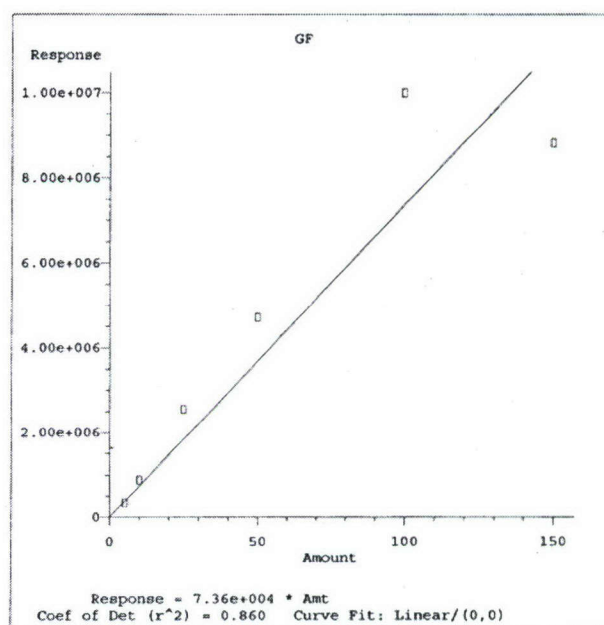
Method Name: C:\HPCHEM\METHODS\ENH\CWA.M
 Calibration Table Last Updated: Fri Feb 09 17:07:38 2007

Figure A-3. GB Standard Curve



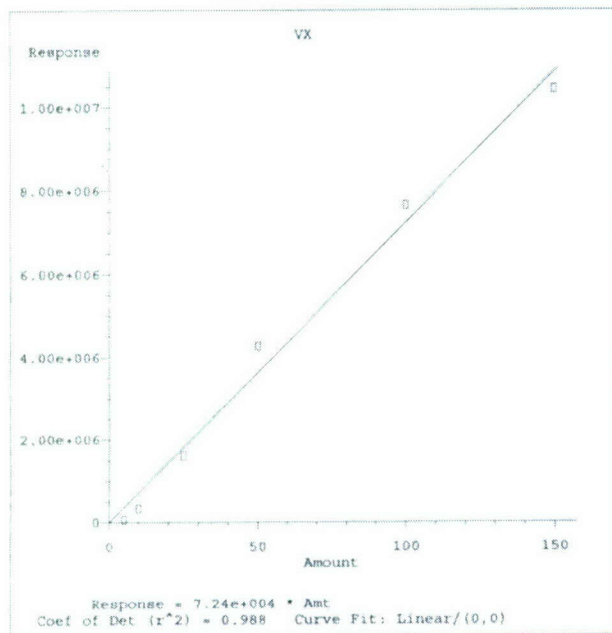
Method Name: C:\HPCHEM\METHODS\ENH\CWA.M
 Calibration Table Last Updated: Fri Feb 09 17:07:38 2007

Figure A-4. GD Standard Curve



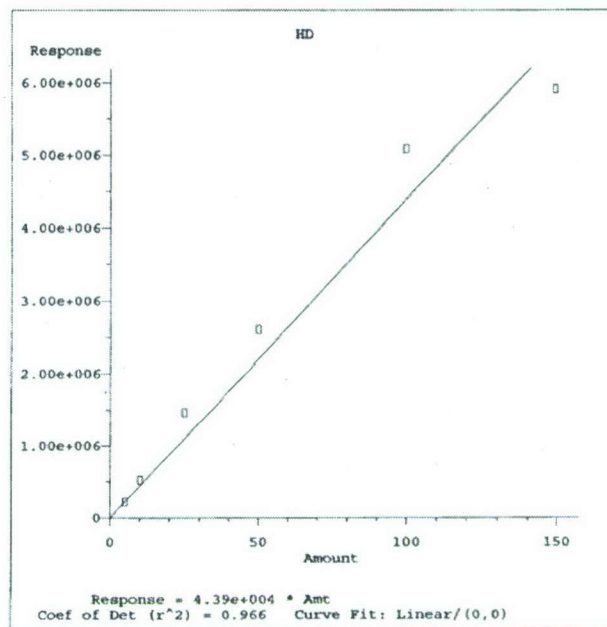
Method Name: C:\HPCHEM\METHODS\ENH\CWA.M
 Calibration Table Last Updated: Fri Feb 09 17:07:38 2007

Figure A-5. GF Standard Curve



Method Name: C:\HPCHEM\METHODS\ENH\VXCWA.M
 Calibration Table Last Updated: Mon Feb 12 17:57:14 2007

Figure A-6. VX Standard Curve



Method Name: C:\HPCHEM\METHODS\ENH\CWA.M
 Calibration Table Last Updated: Fri Feb 09 17:07:38 2007

Figure A-7. HD Standard Curve